LACT2

cobas®

Lactate Gen.2

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
03183700 190	Lactate Gen.2 100 tests	System-ID 07 6606 2	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
10759350 190	Calibrator f.a.s. (12 x 3 mL)	Code 401	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	Code 401	
12149435 122	Precinorm U plus (10 x 3 mL)	Code 300	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	Code 300	
12149443 122	Precipath U plus (10 x 3 mL)	Code 301	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	Code 301	
10171743 122	Precinorm U (20 x 5 mL)	Code 300	
10171735 122	Precinorm U (4 x 5 mL)	Code 300	
10171778 122	Precipath U (20 x 5 mL)	Code 301	
10171760 122	Precipath U (4 x 5 mL)	Code 301	
05117003 190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 391	
05947626 190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 391	
05947626 160	PreciControl ClinChem Multi 1 (4 x 5 mL, for USA)	Code 391	
05117216 190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 392	
05947774 190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 392	
05947774 160	PreciControl ClinChem Multi 2 (4 x 5 mL, for USA)	Code 392	
04489357 190	Diluent NaCl 9 % (50 mL)	System-ID 07 6869 3	

English

System information

For **cobas c** 311/501 analyzers:

LACT2: ACN 040

SLAC2: ACN 047 (STAT, reaction time: 7)

For **cobas c** 502 analyzer:

LACT2: ACN 8040

SLAC2: ACN 8047 (STAT, reaction time: 7)

Intended use

In vitro test for the quantitative determination of lactate in human plasma and CSF on Roche/Hitachi **cobas c** systems.

Summary

Anaerobic glycolysis markedly increases blood lactate and causes some increase in pyruvate levels, especially with prolonged exercise. The common cause for increased blood lactate and pyruvate is anoxia resulting from such conditions as shock, pneumonia and congestive heart failure. Lactic acidosis may also occur in renal failure and leukemia. Thiamine deficiency and diabetic ketoacidosis are associated with increased levels of lactate and pyruvate.

Lactate levels in cerebrospinal fluid are increased in bacterial meningitis. Increased CSF levels also occur in hypocapnia, hydrocephalus, brain abscesses, cerebral ischemia and any clinical condition associated with reduced oxygenation of the brain and/or increased intracranial pressure.

Lactate measurements that evaluate the acid-base status are used in the diagnosis and treatment of lactic acidosis (abnormally high acidity in the blood).

In recent years, enzymatic methods for the determination of lactate have gained favor over colorimetric and titrimetric methods. Enzymatic methods are generally simple and provide greater specificity, accuracy, and reproducibility.

The first enzymatic method described for the determination of lactate was based on the transfer of hydrogen from lactate to potassium ferricyanide by lactate dehydrogenase. However, the procedure was cumbersome and did not receive wide acceptance.

Subsequent methods involved the UV measurement of the formation of NADH. In 1974, Gutmann and Wahlefeld[1] described a lactate procedure that measures the NADH formed by the oxidation of lactate catalyzed by LD, using hydrazine as a trapping agent for pyruvate. A method described by Noll[2] is also based on the catalytic action of LD but includes ALT in the reaction mixture to more rapidly remove the pyruvate formed from the conversion of lactate.

The method presented here uses an enzymatic reaction to convert lactate to pyruvate. The hydrogen peroxide produced by this reaction is then used in an enzymatic reaction to generate a colored dye.[3,4] This method offers longer reagent stability than the previous UV enzymatic methods.

Test principle

Colorimetric assay.

L-lactate is oxidized to pyruvate by the specific enzyme lactate oxidase (LOD). Peroxidase (POD) is used to generate a colored dye using the hydrogen peroxide generated in the first reaction.[3,4]

$$\text{L-lactate} + O_2 \xrightarrow{\text{LOD}} \text{pyruvate} + H_2O_2$$

$$2\,H_2O_2 + \text{H donor} + \text{4-AAP} \xrightarrow{\text{POD}} \text{chromogen} + 2\,H_2O$$

The intensity of the color formed is directly proportional to the L-lactate concentration. It is determined by measuring the increase in absorbance.

Reagents - working solutions

R1 Hydrogen donor: 1.75 mmol/L; ascorbate oxidase (cucumber): 501 µkat/L; buffers; preservatives

R2 4-Aminoantipyrine: 5 mmol/L; lactate oxidase (microbial): 251 µkat/L; peroxidase (horseradish): 401 µkat/L; buffers; preservatives

R1 is in position B and R2 is in position C.

Precautions and warnings

For in vitro diagnostic use.
Exercise the normal precautions required for handling all laboratory reagents.
Disposal of all waste material should be in accordance with local guidelines.
Safety data sheet available for professional user on request.

For USA: For prescription use only.

Reagent handling

Ready for use

Storage and stability

LACT2

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks
Diluent NaCl 9 %	
Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks

Specimen collection and preparation

For specimen collection and preparation only use suitable tubes or collection containers.

Only the specimens listed below were tested and found acceptable.
Serum: **Do not use serum specimens.**
Plasma: Na-fluoride/K-oxalate and Na-fluoride/Na-heparin plasma. Centrifuge within 15 minutes of collecting the specimen.
CSF: May be used as obtained.

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Centrifuge samples containing precipitates before performing the assay.

Note

1. The lactate level increases rapidly with physical exercise. The time required for return to normal lactate values depends on the physical fitness of the subject. 30 minutes at rest is usually sufficient for this purpose.
2. Blood samples should be drawn from a stasis-free vein. However, minimal hemostasis (less than 30 seconds) will not affect lactate levels. Avoid the use of a tourniquet, if possible.[5]
3. Glycolysis in blood samples can rapidly increase lactate levels. Cells contribute to the glycolysis and their quick removal is essential for accurate lactate analysis.[6] Heparinized plasma is acceptable, but precautions must be taken to retard glycolysis by keeping the whole blood on ice and then separating the plasma from the cells within 15 minutes of collection.

Stability in plasma (separated):[7]	8 hours at 15-25 °C
	14 days at 2-8 °C
Stability in plasma (heparinized):[8]	38 days at -20 °C
Stability in CSF:[9]	3 hours at 15-25 °C
	24 hours at 2-8 °C
	2 months at (-15)-(-25) °C

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

- See “Order information” section
- General laboratory equipment

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator’s manual for analyzer-specific assay instructions.

The performance of applications not validated by Roche is not warranted and must be defined by the user.

Application for plasma and CSF

cobas c 311 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 6-31 (STAT 7 / 6-31)		
Wavelength (sub/main)	700/660 nm		
Reaction direction	Increase		
Units	mmol/L (mg/dL, mg/L)		
Reagent pipetting		Diluent (H_2O)	
R1	125 µL	20 µL	
R2	25 µL	20 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	-	-
Decreased	2 µL	15 µL	135 µL
Increased	2 µL	-	-

cobas c 501 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 10-47 (STAT 7 / 10-47)		
Wavelength (sub/main)	700/660 nm		
Reaction direction	Increase		
Units	mmol/L (mg/dL, mg/L)		
Reagent pipetting		Diluent (H_2O)	
R1	125 µL	20 µL	
R2	25 µL	20 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	-	-
Decreased	2 µL	15 µL	135 µL
Increased	2 µL	-	-

cobas c 502 **test definition**

Assay type	2-Point End		
Reaction time / Assay points	10 / 10-47 (STAT 7 / 10-47)		
Wavelength (sub/main)	700/660 nm		
Reaction direction	Increase		
Units	mmol/L (mg/dL, mg/L)		
Reagent pipetting		Diluent (H_2O)	
R1	125 µL	20 µL	
R2	25 µL	20 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	-	-
Decreased	2 µL	15 µL	135 µL
Increased	4 µL	-	-

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	2-point calibration - after reagent lot change - as required following quality control procedures

Traceability: This method has been standardized against a primary standard.

Quality control

For quality control, use control materials as listed in the "Order information" section.

In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

Calculation

Roche/Hitachi **cobas c** systems automatically calculate the analyte concentration of each sample.

Conversion factors:	mmol/L x 9.009 = mg/dL
	mmol/L x 90.09 = mg/L
	mg/dL x 0.111 = mmol/L

Limitations - interference

Criterion: Recovery within ± 10 % of initial value at a lactate concentration of 2.2 mmol/L (19.8 mg/dL).

Plasma

Icterus:[10] No significant interference up to an I index of 28 for conjugated and 60 for unconjugated bilirubin (approximate conjugated bilirubin concentration: 479 µmol/L or 28 mg/dL; approximate unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[10] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[10] No significant interference up to an L index of 1500. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Highly turbid and grossly lipemic samples may cause Abs. flags.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[11,12]

Acetaminophen intoxications are frequently treated with N-Acetylcysteine. N-Acetylcysteine at a plasma concentration above 1497 mg/L and the Acetaminophen metabolite N-acetyl-p-benzoquinone imine (NAPQI) independently may cause falsely low results.

Venipuncture should be performed prior to the administration of Metamizole. Venipuncture immediately after or during the administration of Metamizole may lead to falsely low results. A significant interference may occur at any plasma Metamizole concentration.

Exception: Calcium dobesilate causes artificially low lactate results.

Glycolate, a metabolite of ethylene glycol, causes a positive interference which is variable from lot to lot of reagent. Dicynone (Etamsylate) at therapeutic concentrations may lead to false-low results.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[13]

CSF

No known interference.

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

ACTION REQUIRED

Special Wash Programming: The use of special wash steps is mandatory when certain test combinations are run together on Roche/Hitachi **cobas c** systems. The latest version of the carry-over evasion list can be found with the NaOHD-SMS-SmpCln1+2-SCCS Method Sheets. For further instructions refer to the operator's manual. **cobas c** 502 analyzer: All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is not required.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

0.2-15.5 mmol/L (1.8-140 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:10 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 10.

Lower limits of measurement

Lower detection limit of the test

0.2 mmol/L (1.8 mg/dL)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values

Plasma:	0.5-2.2 mmol/L	(4.5-19.8 mg/dL)	venous[5]
CSF:	1.1-6.7 mmol/L	(10-60 mg/dL)	neonate[5]
	1.1-4.4 mmol/L	(10-40 mg/dL)	3-10 days old
	1.1-2.8 mmol/L	(10-25 mg/dL)	> 10 days old
	1.1-2.4 mmol/L	(10-22 mg/dL)	adult

Roche has not evaluated reference ranges in a pediatric population.

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Specific performance data

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined using human samples and controls in an internal protocol with repeatability (n = 21) and intermediate precision (3 aliquots per run, 1 run per day, 21 days). The following results were obtained:

Plasma

Repeatability	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
Precinorm U	1.70 (15.3)	0.02 (0.2)	1.2
Precipath U	3.24 (29.2)	0.03 (0.3)	1.1
Plasma 1	1.51 (13.6)	0.02 (0.2)	1.3
Plasma 2	2.11 (19.0)	0.02 (0.2)	1.0
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
Precinorm U	1.67 (14.2)	0.03 (0.3)	1.8
Precipath U	3.21 (28.9)	0.05 (0.5)	1.7
Plasma 3	2.38 (21.4)	0.04 (0.4)	1.6
Plasma 4	9.56 (86.1)	0.09 (0.8)	0.9

CSF

Repeatability	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
CSF Control I	1.53 (13.8)	0.03 (0.3)	2.0
CSF Control II	3.95 (35.6)	0.09 (0.8)	2.3
CSF 1	1.85 (16.7)	0.04 (0.4)	2.0
CSF 2	1.33 (12.0)	0.03 (0.3)	2.3
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
CSF Control I	1.53 (13.8)	0.04 (0.4)	2.8
CSF Control II	3.89 (35.0)	0.08 (0.7)	2.1

CSF 3	1.71 (15.4)	0.06 (0.5)	3.3
CSF 4	2.57 (23.2)	0.05 (0.5)	2.1

Method comparison

Lactate values for human plasma and human CSF samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) were compared with those determined using the corresponding reagent on a Roche/Hitachi 917 analyzer (x).

Plasma

Sample size (n) = 69

Passing/Bablok[14]	Linear regression
y = 0.985x + 0.025 mmol/L	y = 0.977x + 0.043 mmol/L
τ = 0.982	r = 1.000

The sample concentrations were between 0.640 and 13.9 mmol/L (5.77 and 125 mg/dL).

CSF

Sample size (n) = 81

Passing/Bablok[14]	Linear regression
y = 1.015x + 0.005 mmol/L	y = 1.010x + 0.015 mmol/L
τ = 0.957	r = 1.000

The sample concentrations were between 0.250 and 9.26 mmol/L (2.25 and 83.4 mg/dL).

References

1 Gutmann I, Wahlefeld A. Methods of Enzymatic Analysis. 2nd ed. Bergmeyer HU, ed. New York, NY: Academic Press Inc 1974:1464.

2 Noll F. Methods of Enzymatic Analysis. 2nd ed. Bergmeyer HU (ed), New York, NY: Academic Press Inc 1974;1475.

3 Trinder P. Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. Ann Clin Biochem 1969;6:24-27.

4 Barhan D, Trinder P. An improved colour reagent for the determination of blood glucose by the oxidase system. Analyst 97;1972:142.

5 Tietz NW, ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia PA: WB Saunders Company 1995;382-383.

6 Burtis CA, Ashwood ER, (eds.). Tietz Textbook of Clinical Chemistry. 2nd ed. Pa: WB Saunders Co 1994;976.

7 Westgard JO, Lahmeyer BL, Birnbaum ML. Use of the Du Pont "automatic clinical analyzer" in direct determination of lactic acid in plasma stabilized with sodium fluoride. Clin Chem 1972;18(11):1334-1338.

8 Nelson SR and Kugler KK. Comparison of Lactate Levels in Acid-Treated and Untreated Blood and Spinal Fluid. Biochemical Medicine 1969;2(4):325-332.

9 Kleine TO. Nervensysteme. In: Greiling H, Gressner AM, eds. Lehrbuch der Klinischen Chemie und Pathobiochemie Stuttgart: Schattauer 1987;859-893.

10 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

11 Breuer J. Report on the Symposium “Drug effects in Clinical Chemistry Methods”. Eur J Clin Chem Clin Biochem 1996;34:385-386.

12 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

13 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

14 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard.

CONTENT	Contents of kit
→	Volume after reconstitution or mixing
GTIN	Global Trade Item Number

FOR US CUSTOMERS ONLY: LIMITED WARRANTY

Roche Diagnostics warrants that this product will meet the specifications stated in the labeling when used in accordance with such labeling and will be free from defects in material and workmanship until the expiration date printed on the label. THIS LIMITED WARRANTY IS IN LIEU OF ANY OTHER WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR PARTICULAR PURPOSE. IN NO EVENT SHALL ROCHE DIAGNOSTICS BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

CE

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com

Distribution in USA by:
Roche Diagnostics, Indianapolis, IN
US Customer Technical Support 1-800-428-2336